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# A new species of *Parapsectrocladius* Cranston (Diptera: Chironomidae: Orthocladiinae) from Patagonia, Argentina

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## Abstract

*Parapsectrocladius setosus* **sp. n.** is described and figured as pupa and male imago, based on material collected in Patagonia, Argentina. The male of the new species groups with *P. escondido* Cranston & Añón Suárez by having the anal point tapering triangular; inferior volsella simple, rounded, without any dorsal ridge and apically blunt. It can be distinguished from *P. escondido* by the possession of strong setae on the inner margin of inferior volsella. New material of *Parapsectrocladius* belonging from several localities allows us to emend the generic diagnosis of the adult male, female and larva of *Parapsectrocladius*, discuss its phylogenetic position and to expand the geographic distribution of *P. acuminatus* Cranston, *P. escondido* and *P. reissi*. Cranston.

Key words: Parapsectrocladius, Orthocladiinae, Phylogeny, Patagonia

## Introduction

The knowledge of the Patagonian Chironomidae diversity has its cornerstone in the work of F. W. Edwards, published in 1931 as the result of the study of specimens collected by him during an expedition in the years 1926–7. Amongst taxa described by Edwards was *Cardiocladius acuminatus*. As the author described in its diagnosis, this species possesses the fourth tarsomere cordiform, as in all the European *Cardiocladius* Kieffer, but differs from them by the stout hairy anal point and the non-pruinose thorax. Particularly the stout hairy anal point led Brundin (1956) to doubt the placement of *acuminatus* in *Cardiocladius*. After the examination of the type of this species, Halvorsen (1988) proposed that the species could be best placed in the genus *Paratrissocladius* Zavřel until additional knowledge from the other life history stages became available. The information provided by the immature stages would not wait long. A reared male together with additional immature and adult specimens from southern Argentina and Chile allowed Cranston (2000) to erect the genus *Parapsectrocladius* for *P. acuminatus* (Edwards) and describe three new species: *P. longistylus* Cranston, *P. reissi* Cranston, and *P. escondido* Cranston & Añón Suárez. The cladistic analysis performed in that study placed *Psectrocladius* Kieffer as the sister genus of *Parapsectrocladius*.

The genus *Parapsectrocladius* is endemic to the Subantarctic forest of Argentina and Chile and lives in lentic environments, except for *P. acuminatus* whose immatures were reported living in running waters. Cranston (2000) described *P. longistylus* as the only species with a trans-Andean distribution, *P. acuminatus* and *P. reissi* occurring in Chile and *P. escondido* occurring in Argentina. The species *P. acuminatus* was cited later from Argentina by Donato *et al.* (2008) from lentic and lotic environments.

The species *P. escondido* is known in all the life stages based on rearing of larva to adult. The association between larva, pupa and adult of *P. acuminatus* was obtained by mass rearing and the female is unknown. The associated larva, pupa and female of *P. reissi* and the association to the pupa and female of *P. longistylus* were based on co-occurrence of the development stages at their type localities.

Concerning this last species, the rearing of two pupae from Nahuel Huapi National Park (Bariloche, Argentina)

shows that *P. longistylus*' pupa should be placed in the new species described in the present paper. Besides, the availability of new material of *Parapsectrocladius* from several localities allows us to emend the generic diagnosis of the adult male, female and larva of *Parapsectrocladius*, discuss its phylogenetic position and to expand the geographic distributions of *P. acuminatus*, *P. escondido* and *P. reissi*.

# Material and methods

Microscope slides were prepared by clearing specimens with 10% KOH; neutralization with glacial acetic acid; dehydration in 80%, 96% and 100% ethanol and mounting in Canada Balsam. Morphological terminology and measurement standards follow Sæther (1980); the values are rounded off to the nearest 5 and are in µm except when otherwise stated; measurements are given as ranges followed by the measurements of the holotype in square brackets. The type material is deposited in the collection of the Museo de La Plata (La Plata, Argentina, MLP), the Natural History Museum (London, UK, NHM) and Zoologische Staatssammlung München (München, Germany, ZSM).

The description of the new species and the re-examination of the diagnostic characters for the genus allow emending its diagnosis. Of particular interest is the presence of the taeniate lateral setae on segments V to VIII in the pupa of *Parapsectrocladius*. The examination of the pupae deposited in MLP showed that all lateral setae on segments V–VIII are simple (Fig. 5). Martin Spies, who kindly examined the pupae of *P. longistylus* deposited in ZSM, observed the same character condition.

As some of the emended characters are taxonomically important and could have phylogenetic implications, two cladistic analyses were performed. One of them was carried out using the data matrix compiled by Cranston (2000) in which the author selected the terminal taxa to evaluate the various hypotheses of relationships of Parapsectrocladius. Two characters were modified in the data matrix, pupal lateral setae on VI (coded with the character state simple) and acrostichals (coded as polymorphic with the character states continuous and absent, since in the new species the acrostichals are absent). The other cladistic analysis was performed using the data matrix and the character list of Mendes et al. (2004) but using genera as terminals and adding the genera Parapsectrocladius, Cardiocladius, and Paratrissocladius. Characters 28, 31, 36, 40, 51, 52, 53, 54, 55, 56, 57, 58, 67, 68, 69, 70, 71, 72, 73, 77 and 125 were not taken into account since the information they provide at this taxonomic level is superfluous (Mendes et al. 2004 assessed the phylogenetic relationships of 38 genera and 25 species entered separately for Antillocladius Kieffer, Compterosmittia Sæther and an unassigned species). The genus Prodiamesa Kieffer was used to root and polarize the tree. Both analyses were conducted in the program TNT, version 1.1 (Goloboff et al., 2008) applying maximum parsimony as optimality criteria and using a traditional search of 1000 random addition sequences plus tree bisection and reconnection (TBR) with the addition of a swapping of the trees obtained with TBR. Character support was estimated with absolute and relative Bremer support values saving up to 6 steps longer suboptimal trees obtained with branch swapping.

# Results

# Taxonomy

# Parapsectrocladius emended diagnosis

The following characters are new with respect to the original description:

**Male**. Wing length 1.54–2.84 mm. *Head*: Antenna with 12–13 flagellomeres. AR 0.94–1.66. *Thorax*: Acrostichals restricted to sector 1/3 from anterior or lacking. *Wing*: Membrane with fine to moderate punctation, without macrotrichia. R with 9–18,  $R_1$  with 0–8 setae. Squama with 9–28 setae. VR between 1.12–1.22. Legs with 3–11 sensilla chaetica in basal 1/10 of tarsomere 1 of midleg.

**Female**. A.R. 0.39–0.55. Wing vein R with 17–20,  $R_1$  with 7–8, and  $R_{4+5}$  with 5–8 setae. VR 1.15–1.23; squama with 16–24 setae. Legs with 6–13 sensilla chaetica in apical quarter of tarsomere 1 of midleg and 2–6 on tarsomere 1 of hindleg.

**Larva**. Fourth-instar larva medium-sized, 2.98-7.2 mm long. Antenna, AR 1.2–2.32. Mentum comprising paired median teeth and 1<sup>st</sup> laterals somewhat appressed or not. Ventromental plate extending from mid first lateral teeth or second teeth, narrow and not extending to outermost mental tooth.

# Key to the male imagines of *Parapsectrocladius* Cranston (modified from Cranston, 2000)

1.	Base of anal point evenly tapering anteriorly to merge with tergite. Inferior volsella rounded and without any dorsal ridge2
-	Base of anal point flanked by setose protuberances. Inferior volsella with substantial dorsally-directed sclerotised ridge 4
2.	Anal point rounded, rather densely setose to apex; inferior volsella distinctly bilobed, dorsal lobe rounded apically
-	Anal point tapering triangular; inferior volsella simple, apically bluntly triangular
3.	Inner margin of inferior volsella with fine setae (Fig 2B of Cranston 2000) P. escondido Cranston et Añón Suárez
-	Inner margin of inferior volsella with strong setae (Fig. 3)
4.	Gonostylus more normal length, not extending to anal point apex when reflexed P. reissi Cranston
-	Gonostylus very long, extending to anal point base when reflexed P. longistylus Cranston

# Parapsectrocladius setosus sp. nov.

**Type material.** ARGENTINA: Holotype male, Río Negro, P. N. Nahuel Huapi, C° Catedral, Laguna Toncek, 41°11'54.2"S– 71°29'12"W, 1747 m a.s.l., 20.i.2007, sweep net, M. Donato col. (MLP). Paratypes: male, same data as holotype; 2 males, same data as holotype except A. Garré and F. Montes de Oca cols. (NHM and ZSM); 2 males with pupal exuviae, same data as holotype except A. Garré and F. Montes de Oca cols., drift net (MLP and NHM); 1 male, Río Negro, Meseta de Somuncurá, Laguna Puesto de Lata, Paraje Chasicó, Estancia Jaguar Musso, 41°09'18.6"S– 67°34'40.3"W, 1.055 m a.s.l., M. Donato col. (MLP).

Etymology. Named after the strong and numerous setae in the inner margin of the inferior volsella.

**Diagnosis.** The male of *Parapsectrocladius setosus* is clearly differentiated from the rest of the species of the genus except *P. escondido* by having the anal point tapering triangular without setose protuberances at base and the inferior volsella simple and apically bluntly triangular. It can be separated from *P. escondido* by having the inner margin of the inferior volsella with strong setae. The pupa of *P. setosus* is separated from the rest of the members of the genus by having the thoracic armament comprising long blunt spines (>30pm), thoracic horn covered with fine spinules and with a basal swelling, median spine patches of tergites IV–VI weakly defined and apex of anal lobe with small tubercles.

**Male** (n = 7). Total length 3.86–5.40, [4.98] (6) mm. Wing length 2.22–2.84, [2.74] (5) mm. Total length/wing length 1.74–2.03, [1.82] (5). Wing length/length of profemur 2.37–2.52, [2.43] (5). Coloration (specimen preserved in alcohol) uniformly brown; thorax somewhat light brown with vittae, postnotum and preepisternum darker; legs light brown.

Head. Antennae with 12–13, 13 (6) flagellomeres; AR 1.2–1.5, [1.41] (5), ultimate flagellomere 489–584, [568] (5) long. Temporal setae 8, divided in 1, [1] inner verticals, 1–2, [1] outer verticals, and 3–5, [4] postorbitals. Clypeus with 18–28, [28] (6) setae. Tentorium 220–254, [230] (6) long and 46–100, [48] (6) wide. Palpomere lengths (1–5) (5): 40–60, [40]; 50–80, [62]; 100–150, [124]; 118–148, [130]; 172–224, [186] (4).

Thorax. Antepronotum with 9-15, [11] (5) lateral setae. Acrostichals absent. Dorsocentrals 18-22, [22] (4); prealars 6-9, [8] (5); supraalar 1, [1] (5). Scutellum with 9-18, [18] (5) setae.

Wing. VR 1.13–1.2, [1.16] (5). C extension 20–78, [20] (5) long. Brachiolum with 2, [2] (5) seta. R with 10–12, [10] (4);  $R_1$  with 0–1, [0] (5);  $R_{4+5}$  with 0–2, [2] (5). Squama with 12–28, [23] (5) setae.

Legs. Spur of front tibia 60-154, [62] long; spurs of mid tibia 24-40, [40] and 32-44, [44]; of hind tibia: 30-42, [42] (6) and 76-98, [90] long. Width at apex of front tibia 60-88, [70]; of mid tibia 70-154, [74]; of hind tibia 80-106, [84]. sensilla chaetica of tarsomere 1 of midleg 3-6, [4] (6). Comb with 11-13, [11] (6) setae. Lengths and proportions of legs in Table 1.

Hypopygium (Figs.1–3). Tergum IX with 18 setae, divided in 4–7, [4] (6) small setae and 7–11, [7] (6) big setae. Laterosternite IX with 3–6, [3] (5) setae. Anal point 162–196, [162] (6) long, width at base 30–60, [44] (6), and with 14–24, [16] (6) setae. Phallapodeme 130–162, [162] (5) long; transverse sternapodeme 94–114, [114] (5) long. Virga represented by a broad and diffuse area of fine spinules. Gonocoxite 362–462, [450] long; length from

base of gonocoxite to apex of inferior volsella/length of gonocoxite 0.53–0.55, [0.53]. Inferior volsella subtriangular with strong setae on inner margin (Fig. 3). Gonostylus 168–202, [180] long; megaseta 18–26, [20] (6) long. HR 2.15–2.5, [2.5]; HV 2.30–2.77, [2.77] (6).



FIGURES 1–3. *Parapsectrocladius setosus* sp. nov., male. 1, Hypopygium, dorsal aspect; 2, Hypopygium with tergite IX removed, dorsal aspect to the left, ventral aspect to the right; 3, Inferior volsella.

**Pupa** (n= 2) (Figs. 4–5). The pupae examined fit the description by Cranston (2000) of *P. longistylus* except for the presence of an anteromedian patch of spines present on T III (Fig. 4). In the drawing of the pupal abdomen (Fig. 4A of Cranston, 2000) the tergites IV–VI have an anteriomedian patch of spines.

# Female and larva. Unknown.

Distribution and biology. The species is known from Chile and Argentina. The altitude range is from about

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1000 to 1.700 m a.s.l. Collection sites show that this species prefers standing water environments. The geographic distribution of the genus is extended eastward in Patagonia since *P. setosus* was collected in the Somuncurá Plateau, a vast basalt plateau of over 25,000 km<sup>2</sup> located in north eastern Patagonia, with an average altitude of 1000 m a.s.l. The terrestrial vegetation is predominantly herbaceous and shrub forms (León *et al.*, 1998). This plateau would not have suffered the Patagonian glaciations and subsequent marine ingressions, thereby acting as a refuge and dispersal center (Ringuelet, 1975). The Somuncurá Plateau is also an area of endemism, with numerous endemic species such as the fish *Gymnocharacinus bergi* Steindachner (Menni & Gomez, 1995), the frogs *Atelognathus reverberii* (Cei) and *Pleurodema somuncurensis* (Cei) (Cei, 1980), the scorpions *Urophonius somuncura* Acosta (Acosta, 2003) and *Bothriurus ceii* Ojanguren-Affilastro (Ojanguren-Affilastro, 2007), among others.



FIGURES 4-5. Parapsectrocladius setosus sp. nov., pupa. 4, Tergite III; 5, Tergite VIII.

**TABLE 1.** Lengths (in µm) and proportions of legs of *Parapsectrocladius setosus* **sp. n.** (male, n = 5). Measurements of the holotype in square brackets. Abbreviations: Femur (fe); Tibia (ti); Tarsomeres 1–5 (ta<sub>1-5</sub>); Leg Ratio (LR), ratio of metatarsus to tibia; «Beinverhältnisse» (BV), combined length of femur, tibia, and basitarsus divided by combined length of tarsomeres 2–5; «Schenkel-Scheine-verhältniss» (SV), ratio of femur plus tibia to metatarsus.

	fe	ti	ta <sub>1</sub>	ta <sub>2</sub>	ta <sub>3</sub>
P <sub>1</sub>	939–1128 [1128]	1144–1452 [1452]	868–1089 [1089]	473–592 [592]	276–331 [331]
$P_2$	963–1160 [1144]	978–1318 [1302]	544-679 [663]	300–379 [379]	189–237 [221]
$P_3$	1034–1262 [1215]	1255–1554 [1539]	773–963 [947]	418–513 [497]	205–252 [237]
	ta <sub>4</sub>	ta <sub>5</sub>	LR	BV	SV
P <sub>1</sub>	150–181 [174]	166–205 [189]	0.73–0.77 [0.75]	2.77–2.85 [2.85]	2.36–2.48 [2.37]
$P_2$	126–158 [142]	142–197 [174]	0.50-0.65 [0.51]	2.81–3.44 [3.40]	3.08–3.78 [3.69]
$P_3$	142–174 [150]	150–189 [174]	0.62–0.63 [0.62]	3.29–3.57 [3.50]	2.89–2.96 [2.91]

## Parapsectrocladius acuminatus (Edwards)

#### Cardiocladius acuminatus Edwards 1931: 275.

Paratrissocladius acuminatus; Halvorsen 1988 (comb. n., redescription of male); Spies & Reiss 1996 (catalog).

Parapsectrocladius acuminatus; Cranston 2000 (comb. n., description of larva and pupa); Donato et al. 2008 (checklist, new record), Ashe & O'Oconnor 2012 (catalog).

**Material examined.** ARGENTINA: 6 males, Río Negro, P. N. Nahuel Huapi, Manso medio, La Cantera, 41°21'16"S–71°42'27.3"W, 764 m a.s.l., 15-i/2-ii-2007, Malaise trap, Garré-Montes de Oca cols. (MLP); 1male, Río Negro, P. N. Nahuel Huapi, río Ñirihuau, 41°17'35.1"S–71°14'26.3"W, 1044 m a.s.l., 15-ii/28-ii-2007, Malaise trap, Garré-Montes de Oca cols. (MLP); 2 males, Tierra del Fuego, Río Moat y Ruta Comp. J, 54°58'03.9"S–66°44'35.7"W, 2-xii-2008, Malaise trap, M. Donato & G. Spinelli cols. (MLP).

**Male** (n = 9). Total length 2.80–3.82 mm. Wing length 1.54–2.06 mm. Total length/wing length 1.63–2.05. Wing length/length of profemur 2.37–2.58.

Head. AR 0.98–1.26, ultimate flagellomere 371–481 long. Temporal setae 5–9, divided in 1 inner vertical and 4–8 outer verticals. Clypeus with 6–14 setae. Tentorium 164–208 long and 30–60 wide. Palpomere lengths (1–5): 44–62; 42–70; 112–150; 108–140; 160–196.

Thorax. Antepronotum with 6–8 lateral setae. Acrostichals 5–10 (6). Dorsocentrals 7–13; prealars 2–4; supraalar 0–1 (7). Scutellum with 5–11 setae.

Wing. VR 1.14–1.22. C extension 0–36 (8) long. Brachiolum with 1–2 seta. R with 9–18;  $R_1$  with 3–8 setae. Squama with 10–20 setae.

Legs. Spur of front tibia 48–90 (8) long; spurs of mid tibia 18–28 and 24–32; of hind tibia: 18–30 and 56–99 long. Width at apex of front tibia 42–56; of mid tibia 40–58; of hind tibia 48–70. Sensilla chaetica of tarsomere 1 of midleg 4–11. Comb with 9–11 setae. Lengths and proportions of legs in Table 2.

**TABLE 2.** Lengths (in  $\mu$ m) and proportions of legs of *Parapsectrocladius acuminatus* (Edwards) (male, n = 9). For abbreviation explanations see Table 1.

	Fe	ti	ta <sub>1</sub>	ta <sub>2</sub>	ta <sub>3</sub>
P <sub>1</sub>	647–797	797–947	615–789	323–434	197–252
$P_2$	655-828	742–884	379–497	197–276	110–158
P <sub>3</sub>	679–907	773–1089	418–718 (8)	221-402 (8)	126–189 (8)
	ta <sub>4</sub>	ta <sub>5</sub>	LR	BV	SV
P <sub>1</sub>	95–118	103–126	0.77–0.83	2.72–2.87	2.21–2.35
$P_2$	71–103	95–134	0.51–0.56	3.28–3.75	3.44–3.83
$P_3$	79–118 (8)	87–142 (8)	0.54-0.66 (8)	3.19-3.65 (8)	2.77-3.55 (8)

Hypopygium. Tergum IX with 6-10 (8) setae. Anal point 80-130 (8) long, width at base 30-44 (6), and with 12-17 (7) setae.Phallapodeme 78–176 long; transverse sternapodeme 66-82 long. Gonocoxite 264-328 long; length from base of gonocoxite to apex of inferior volsella/length of gonocoxite 0.6-0.68. Gonostylus 104-159 long; megaseta 10-18 (8) long. HR 1.97-2.56; HV 2.14-3.05.

**Geographic distribution**. This species was known from the Valdivian forest in Chile (its holotype and the new material studied by Cranston 2000) and the surrounding area of Bariloche in Rio Negro province (Argentina) (Donato *et al.* 2008). New material allows us to extend its distribution to Tierra del Fuego province (Argentina), located in the southernmost distribution of the Subantarctic forest.

## Parapsectrocladius escondido Cranston et Añón Suárez

Parapsectrocladius escondido Cranston et Añón Suárez in Cranston 2001: 440; Donato et al. 2008 (checklist), Ashe & O'Oconnor 2012 (catalog).

Parapsectrocladius escondido Cranston et Añón Suárez in Cranston 2000: 115. Nomen nudum.

**Material examined.** ARGENTINA: Holotype male with larval and pupal exuviae, Río Negro, P. N. Nahuel Huapi, Bariloche, Lago Escondido, 5-i-1998, Añón Suárez col. (MLP #2678); 4 males, Río Negro, P. N. Nahuel Huapi, laguna Los Cántaros, 41°00'20.5"S–71°49'19.5"W, 874 m a.s.l., 19-xii-2007/7-i-2008, Malaise trap, Garré-Montes de Oca cols. (MLP); 4 females, Río Negro, P. N. Nahuel Huapi, mallín La Heladera, 41°00'06.4"S–71°49'40.3"W, 859 m a.s.l., 15-xii-2006, sweep net, Garré-Montes de Oca cols. (MLP).

**Male** (n = 5). Total length 3.60–4.16 (3.60) mm. Wing length 1.80–2.50 (1.80) mm. Total length/wing length 1.64–2.00 (2.00). Wing length/length of profemur 2.51–2.73 (2.51).

Head. AR 1.33–1.66 (1.33), ultimate flagellomere 466–631 (466) long. Temporal setae 7–8 (7), divided in 2–3 (3) inner verticals and 4–5 (4) outer verticals. Clypeus with 7–13 (7). Tentorium 174–212 (174) long and 32–50 (32) wide. Palpomere lengths (1-5): 48–64 (48); 42–68 (42); 122–164 (122); 126–188 (126); 175–236 (175).

Thorax. Antepronotum with 8-14 (14) lateral setae. Acrostichals 7-10 (7) [2]. Dorsocentrals 10-14 (10); prealars 3-4 (3); supraalar 1 (1). Scutellars 7-11 (7).

Wing. VR 1.12–1.20 (1.12). C extension 0–20 (0) long. Brachiolum with 2–3 (2) seta. R with 10–16 (12); remaining veins bare. Squama with 16–23 (16) setae.

Legs. Spur of front tibia 60–70 (60) long; spurs of mid tibia 22–34 (22) and 24–40 (24); of hind tibia: 24–60 (24) and 62–80 (62) long. Width at apex of front tibia 44–56 (44); of mid tibia 44–56 (46); of hind tibia 52–66 (52). Sensilla chaetica of tarsomere 1 of midleg 3–4 (3). Comb with 7–9 [4] setae. Lengths and proportions of legs in Table 3.

	fe	ti	ta <sub>1</sub>	ta <sub>2</sub>	ta <sub>3</sub>
$P_1$	718–963 [718]	892–1191 [892]	734–1018 [734]	387–544 [387]	229–300 [229]
$P_2$	781–1026 [781]	828–1089 [828]	458–647 [458]	237–355 [237]	134–174 [134]
$P_3$	828–1089 [828]	1018–1357 [1018]	671–923 [671]	363–497 [363]	174–229 [174]
	ta <sub>4</sub>	ta <sub>5</sub>	LR	BV	SV
P <sub>1</sub>	ta <sub>4</sub> 103–142 [103]	ta <sub>5</sub> 110–142 [110]	LR 0.82–0.85 [0.82]	BV 2.78–2.98 [2.83]	SV 2.12–2.19 [2.19]
P <sub>1</sub> P <sub>2</sub>	ta <sub>4</sub> 103–142 [103] 95–126 [95]	ta <sub>5</sub> 110–142 [110] 110–126 [110]	LR 0.82–0.85 [0.82] 0.55–0.69 [0.55]	BV 2.78–2.98 [2.83] 3.33–3.73 [3.59]	SV 2.12–2.19 [2.19] 3.05–3.52 [3.52]

**TABLE 3.** Lengths (in  $\mu$ m) and proportions of legs of *Parapsectrocladius escondido* Cranston & Añón Suárez (male, *n* = 5). Measurements of the holotype in square brackets. For abbreviation explanations see Table 1.

Hypopygium. Tergum IX with 6-10 (8) setae. Anal point 58–84 [4] long, width at base 36–80 [4], and with 6-9 [4] setae.Phallapodeme 94–146 (94) long; transverse sternapodeme 64–104 (64) long. Gonocoxite 230–296 (230) long; length from base of gonocoxite to apex of inferior volsella/length of gonocoxite 0.65–0.69 (0.65). Gonostylus 95–116 (95) long; megaseta 14–16 [3] long. HR 2.40–2.74 (2.42); HV 3.53–3.80 (3.79).

**Female** (n=4). Total length 2.64–3.24 mm. Wing length 2.50–2.64 mm. Total length/wing length 1.00–1.30. Wing length/length of profemur 2.88–3.13.

Head. AR 0.39-0.54. Flagellomeres length: 84-90; 44-52; 46-54; 46-56; 92-124. Temporal setae 4-7: including 1–3 inner verticals and 3–4 outer verticals. Clypeus with 11–19 setae. Tentorium 180–200 long, 38-46 wide. Palpomeres length: 44-60; 60-70; 108-139; 140-154; 192-232 (3).

Thorax chaetotaxy. Lateral antepronotals 7–10, Dorsocentrals 11–17, Prealars 3, Supralar 1 (2), Scutelars 10–12.

Wing. VR 1.15–1.23. C extension 0–42 long. Brachiolum with 2 setae. R with 17–20,  $R_1$  7–8,  $R_{4+5}$  5–8. Squama with 16–24 setae.

Legs. Spur of fore tibia 30-40 long; spur of middle tibia 26-34 and 34-48 long; spurs of hind tibia 30-34 and 70-78 long. Width of apex of fore tibia 52-54; of middle tibia 54-60; of hind tibia 60-64. Sensilla chaetica on tarsomere 1 of middle leg 6-13, of hind leg 2-6. Tibial comb with 7-8 setae. Legs measurements as in Table 4.

**TABLE 4.** Lengths (in  $\mu$ m) and proportions of legs of *Parapsectrocladius escondido* Cranston & Añón Suárez (female) (n = 4). For abbreviation explanations see Table 1.

	fe	ti	ta <sub>1</sub>	ta <sub>2</sub>	ta <sub>3</sub>
<b>P</b> <sub>1</sub>	836–868	1057–1152	852–931	450–497	237–276
$P_2$	915–963	947–1034	537–568	276–316	142–166
P <sub>3</sub>	907–986	1089–1278	813-860	418–450	181–197
	ta <sub>4</sub>	ta <sub>5</sub>	LR	BV	SV
<b>P</b> <sub>1</sub>	126–134	134–142	0.75–0.84	2.82-2.99	2.08–2.32
P <sub>2</sub>	110-118	118–134	0.53–0.57	3.42-3.71	3.38–3.61
P <sub>3</sub>	118–126	134	0.64–0.79	3.3–3.46	2.32–2.75

Abdomen. Number of setae on sternite VIII 4–9, Gonocoxite IX with 5–6 long setae and 2–7 small setae. Cercus 122–160 long. Seminal capsules 150–186 long. Notum 230–244 long.

**Remarks.** The new material was found in other standing water environments such as lagoon and mallín. This last environment, is a name from the Patagonian Mapuche native people that refers to a kind of wetland that can be a swampy area or a low area where water accumulates, is characteristic in Patagonia.

## Parapsectrocladius reissi Cranston

#### Parapsectrocladius reissi Cranston 2000: 114.

**Material examined.** ARGENTINA: 7 females, Río Negro, P. N. Nahuel Huapi, Manso medio, La Cantera, 41°21'16"S–71°42'27.3"W, 764 m a.s.l., 15-i/2-ii-2007, Malaise trap, Garré-Montes de Oca cols. (MLP).

**Remarks.** These females were caught in a pond near Manso medio River. This body of standing water could be connected with Manso medio River when the river overflows. The species was first known from Chile and is described in detail by Cranston (2000). These females fit well with Cranston's description pointing out a slight difference in the number of setae in gonocoxite IX, with 4–5 in Chilean specimens and 6–8 setae in the new specimens. These specimens represent the first record of this species from Argentina, thereby becoming a new trans-andean taxon.

## Unplaced specimens of *Parapsectrocladius*

## Parapsectrocladius "Playa Negra"

**Material examined.** ARGENTINA: 1 female, Río Negro, P. N. Nahuel Huapi, Manso medio, mallín Playa Negra, 41°21'26.1"S–71°34'12.6"W, 808 m a.s.l., 26-x-2006, sweep net, Garré-Montes de Oca cols. (MLP).

**Female** (*n*=1). Total length 3.52 mm. Wing length 2.70 mm. Total length/wing length 1.30. Wing length/length of profemur 3.03.

Head. AR 0.50. Flagellomeres length: 92; 48; 58; 60; 128. Temporal setae 6: including 1 inner vertical and 5 outer verticals. Clypeus with 14 setae. Tentorium 192 long, 40 wide. Palpomeres length: 64; 64; 120; 132; 214.

Thorax chaetotaxy. Lateral antepronotals 10, Dorsocentrals 13, Prealars 4, Supralar 1, Scutelars 15.

Wing. VR 1.33. C extension 20 long. Brachiolum with 3 setae. R with 18, R<sub>1</sub> 14, R<sub>4+5</sub> 6. Squama with 36 setae. Legs. Spur of fore tibia broken; spur of middle tibia 30 and 40 long; spurs of hind tibia 32 and 74 long. Width of apex of fore tibia 72; of middle tibia 62; of hind tibia 76. Sensilla chaetica on tarsomere 1 of middle leg 24, of hind leg 25. Tibial comb with 9 setae. Legs measurements as in Table 5.

**TABLE 5.** Lengths (in  $\mu$ m) and proportions of legs of *Parapsectrocladius* "Playa Negra" (female) (n = 1). For abbreviation explanations see Table 1.

	fe	ti	ta <sub>1</sub>	ta <sub>2</sub>	ta <sub>3</sub>
<b>P</b> <sub>1</sub>	892	1136	899	450	276
P <sub>2</sub>	955	1049	552	284	166
P <sub>3</sub>	963	1270	844	410	197
	ta <sub>4</sub>	ta <sub>5</sub>	LR	BV	SV
P <sub>1</sub>	134	142	0.79	2.92	2.25
P <sub>2</sub>	110	118	0.53	3.77	3.63
P <sub>3</sub>	134	134	0.66	3.51	2.64

Genitalia (Fig. 6). Number of setae on sternite VIII 18, Gonocoxite IX with 27 setae. Dorsomesal lobe as in generic diagnosis, apodeme lobe as in Figure 7. Cercus 160 long. Seminal capsules 180 long. Notum 220 long.

**Remarks**. *Parapsectrocladius* "Playa Negra" is very close to *P. reissi* by sharing the undivided tergite IX, gonapophysisVIII, with distinct ventrolateral lobe and 9 strong setae on each side of midline of sternite VIII near gonocoxapodeme. The gonocoxite IX, with 27 setae no longer than 40  $\mu$ m present in the former species is unique among the known females of the genus.



FIGURES 6-7. Parapsectrocladius "Playa Negra". 4, Female genitalia, ventral view; 5, Apodeme lobe.

# Parapsectrocladius sp1

**Material examined.** ARGENTINA: 2 larvae, Río Negro, P. N. Nahuel Huapi, A° Ñireco, desembocadura en lago Nahuel Huapi, 41°07'49"S–71° 17' 20.1" W, 767 m a.s.l., 16-ii-2010, Surber net, M. Mauad col. (MLP).

**Larva** (n=2). Body length 3.64–4.06 mm, head capsule length 323–395. Antenna 56–62, 20–20, 8–6, 4–4, 2–2; AR 1.65–1.94; blade and style unmeasurable. Mandible length 108–130. Mentum (Fig. 8) width 102–116, median mentum width 22–24. Procercus length 30–40, width 16–24, anal seta 226 (1).

# Parapsectrocladius sp2

**Material examined.** ARGENTINA: 1 larva, Río Negro, P. N. Nahuel Huapi, A° Ñireco, desembocadura en lago Nahuel Huapi, 41°07'49"S–71° 17' 20.1" W, 767 m a.s.l., 16-ii-2010, Surber net, M. Mauad col. (MLP); 1 larva, Río Negro, P. N. Nahuel Huapi, A° Ñireco, 41°10'51"S–71° 19' 8.8" W, 892 m a.s.l., 2-xii-2009, Surber net, M. Mauad col. (MLP).

**Larva** (n=2). Body length 2.98–3.48 mm, head capsule length 268–292. Antenna 44–50, 12–14, 3–3, 3–3, 1–2; AR 2.27–2.32; blade and style unmeasurable. Mandible length 74–88. Mentum (Fig. 9) width 80–98, median mentum width 18–20. Procercus length 32, width 20, anal seta 424 (1).



FIGURES 8–9. Mentum. 6, Parapsectrocladius sp1; 7, Parapsectrocladius sp2.

# **Phylogenetic results**

The parsimony analysis was performed using the data matrix compiled by Cranston (2000), some new character settings are here proposed: some characters were treated as additive, others as non-additive; a new codification was added for the simple pupal lateral setae on VI segment, acrostichals were coded as polymorphic. Parsimony analysis produced 3 trees of length 317 (CI= 27, RI= 53). In a tree, *Parapsectrocladius* is sister to the clade *Psectrocladius* (Fig. 10A). This relationship is supported by the antennal segment 3 longer or subequal to segment 4, larval SI seta palmate, one premandibular tooth, presence of sensilla chaetica on hindleg, female apodeme lobe distinct, and female labia microtrichiose. In the other two trees (Fig, 10B and C), *Parapsectrocladius* is ssociated with a clade containing [*Orthocladius (Symposiocladius)* [*Parorthocladius* [[*Eukiefferiella, Tvetenia*] [*Cardiocladius, Lopescladius*]]] and a clade including [*Vivacricotopus* [*Paracricotopus* [[*Rheocricotopus, Psilocricotopus*] [[*Ps. (Allopsectrocladius), Ps. (Mesopsectrocladius)*] [(*Ps. s.str., Ps. Monopsectrocladius)*]]]] respectively. Bremer support values fell below 66% and are not given. From this analysis, one tree agrees with the relationship hypothesis of *Psectrocladius* as a sister group of *Parapsectrocladius* proposed by Cranston (2000).

One of the remaining trees places *Parapsectrocladius* close to *Cardiocladius*, thus approaching to the hypothesis proposed by Edwards (1931).



**FIGURE 10.** Hypothesized cladograms for *Parapsectrocladius* and putative relatives derived using the modified data matrix of Cranston (2000).



**FIGURE 11.** Strict consensus tree obtained from the data matrix modified from Mendes *et al.* (2004) (9 trees of length 796, CI = 0.22, RI = 0.46).

The cladistic analysis carried out using the data matrix modified by Mendes *et al.* (2004) yielded 9 trees of length 796 (CI= 0.22, RI= 0.46); the results are summarized in the strict consensus tree shown in Figure 11. The genus *Psectrocladius* is the sister group of *Parapsectrocladius* sharing the presence of pseudospurs; pupal frontal apotome with warts; pupal tergal armament including elongate, parallel-sided, needle-like spines; larval SI seta palmate.

The cladistic analyses performed in the present study after the emendation of the characters pupal lateral setae on VI simple and acrostichals coded as polymorphic with the character states continuous and absent did not elucidate the phylogenetic relationships of *Parapsectrocladius*. The characters cited by Edwards (1931) and Halvorsen (1988) in their discussions of generic allocation have shown the same problems in these analyses as those considered by Cranston (2000). In the recent Chironomidae phylogenetic hypothesis proposed by Cranston *et*  *al.* (2012) based on molecular evidence, *Parapsectrocladius acuminatus* is the sister group of a clade [*Rheocricotopus* [*Synorthocladius* [*O. (Euorthocladius)* luteipes [*Cricotopus* [*Cricotopus*, *Paratrichocladius*]]]]]. All the morphological-based phylogenies here discussed are constrained by high levels of homoplasy and therefore generating difficulties to recognize characters that may provide unambiguous evidence of relationships. As an exercise, different analyses in both data matrices were performed either using prior weightings in those characters proposed by the authors to support their phylogenetic hypotheses or the application of implied weights, but all results were poorly supported.

Future studies including more taxa together with morphological evidence will better resolve these conflicting relationships.

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